CONTROVERSIES IN TRANSFUSION MEDICINE

Consistency and Proportionality in Policy Decision-Making in Blood Safety: the Case for an All-Apheresis Platelet Supply in Germany

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SUMMARY

Recently, German investigators presented the first mathematical model finding a significant increase in the risk of HIV, HCV, and HBV transmission when pools of 4 whole-blood-derived buffy-coat platelets, rather than 1 single-donor (apheresis) component, are used to provide one platelet dose. Based, in both cases, on mathematical models employing the incidence/window-period method, the relative risk of transmission from pooled versus apheresis platelets (2.2 or 2.75 for HIV, 2.7 or 3.375 for HCV, and 3.2 or 4.0 for HBV, with pools of 4 or 5 concentrates, respectively) is similar to the difference in risk before (versus after) introduction of HIV-1 and HCV RNA screening. The absolute increase in the risk from pools (1 to 2 HIV-, HCV-, or HBV-infectious platelet doses annually) is much smaller than the yield from HIV-1 and HCV RNA screening projected in the 1990s, but it becomes similar to that yield (with up to 88 infectious platelet doses intercepted) when we consider the next transfusion-transmitted pathogen to emerge in the future. Although pathogen reduction (PR) of platelets would eliminate the difference in risk between pooled and apheresis platelets vis-à-vis viral transmission, PR is not ready for implementation because the safety of PR needs to be investigated further. German transfusion guidelines should be revised to indicate the difference in risk associated with pooled versus apheresis platelets, and transition toward an all-apheresis platelet supply should commence. These actions are consistent with and proportionate to the action taken in the 1990s when screening for HIV-1 and HCV RNA was implemented.

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Table 1. Number of transmissions (during the year preceding the introduction of blood-donor screening) of the next “HIV-like” pathogen to emerge in the future, depending on how the 260,000 platelet doses (annually transfused as pools in the US) are obtained†.

<table>
<thead>
<tr>
<th>Type of donor</th>
<th>Presumed mean number of donations per year</th>
<th>Presumed percentage of split apheresis donations</th>
<th>Needed number of donors</th>
<th>Number of transmissions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apheresis</td>
<td>10</td>
<td>100</td>
<td>13,000</td>
<td>26 [a]</td>
</tr>
<tr>
<td>Apheresis</td>
<td>5</td>
<td>100</td>
<td>26,000</td>
<td>26 [b]</td>
</tr>
<tr>
<td>Apheresis</td>
<td>5</td>
<td>50</td>
<td>34,667</td>
<td>26 [c]</td>
</tr>
<tr>
<td>Apheresis</td>
<td>2</td>
<td>0</td>
<td>130,000</td>
<td>26 [d]</td>
</tr>
<tr>
<td>Whole-blood</td>
<td>2</td>
<td>N/A</td>
<td>650,000</td>
<td>130 [e]</td>
</tr>
</tbody>
</table>

† Modified from Vamvakas [3] and based on the assumption that the next “HIV-like” pathogen will attain a prevalence of 1 per 10,000 donors in the year preceding the introduction of blood-donor screening. Regardless of what assumptions are made about the number of donations per year made by apheresis versus whole-blood donors, or the proportion of plateletapheresis collections that are split, if the 260,000 platelet doses currently transfused as pools in the US [4] are transfused as apheresis components in the future, there will be 26 transmissions annually of the next major “HIV-like” pathogen from these 260,000 platelet doses. Conversely, if the 260,000 platelet doses now transfused as pools continue to be transfused as pools, there will be 130 transmissions of the new agent during the year preceding the introduction of blood-donor screening. For more detail, and for a discussion of the assumptions made in the calculations, see Vamvakas [2].

a. 13,000 donors times 10 donations per year times 2 (for splitting every apheresis collection into 2 components) divided by 10,000 (the assumed prevalence of the next “HIV-like” pathogen in US blood donors) equals 26 recipient infections.
b. 26,000 donations times 5 times 2 divided by 10,000 = 26 recipient infections.
c. 34,667 times 5 (i.e., 173,335) plus 86,667 (the one half of the 173,335 collections that are split) divided by 10,000 = 26 recipient infections.
d. 130,000 times 2 divided by 10,000 = 26 recipient infections (option avoiding any adverse effects potentially associated with frequent apheresis, such as citrate toxicity or suppression of platelet counts).
e. 650,000 times 2 divided by 10,000 = 130 recipient infections.

N/A = Not applicable.

vided as a pool of 4 WBD platelet concentrates, increases the risk of HIV transmission by 2.2 (rather than 4) times; the risk of HCV transmission by 2.7 (rather than 4) times; and the risk of HBV transmission by 3.2 (rather than 4) times (Figure 1) [1].

The model built by the German investigators [1] is of great importance, because it puts to rest concerns that the risk of transmission of HIV, HCV, and/or HBV infection might be increased with single-donor platelets compared with platelet pools because apheresis donors donate more often than whole-blood donors and apheresis components are often split. The fear that an infected plateletapheresis donor(s) who donate(s) many times a year and whose donations are split and transfused to multiple recipients can, by infecting so many recipients, lead to an overall increase in the risk of transmission of a transfusion-transmitted infection (TTI) by apheresis (compared with pooled WBD) platelets has also been allayed by others (Table 1) [2,3]. As the German investigators [1] found, such an infected plateletapheresis donor(s) who donate(s) many times a year and whose donations are split would be responsible for a cluster(s) of TTI in space and time, but not for an increase in the overall risk of transmission of a specific TTI secondary to transfusion of single-donor versus pooled platelets. The German investigators concluded that, in the event of an increase in the proportion of therapeutic platelet doses transfused as single-donor (rather than pooled) platelets in Germany, the changes in donor demographic-ics (with respect to age, gender, and population size of place of residence) need to be taken into account when considering the risk of infectious disease transmission. If the next major transfusion-transmitted pathogen to emerge were concentrated in young males from large urban areas, there would probably be no difference in the risk of its transmission between pooled and single-donor platelets because young males from large urban areas (who would be more likely to be infected) are also overrepresented among apheresis (compared with whole-blood) donors. Conversely, if the next agent were concentrated in older residents of small cities and rural areas (or—in the extreme case—in older females residing in small cities and rural areas), the difference in risk between platelet pools and single-donor platelets would be greater than the 4-fold (for pools of 4) or 5-fold (for pools of 5) difference in risk expected from the difference in number of donor exposures [1].

The caution that the German investigators [1] advocate (before we consider making changes to the composition of the donor population) is laudable because—although transfusion-related mortality is currently very low [5]—a novel and fatal TTI could emerge and reproduce the HIV catastrophe of the 1980s [2,3,6,7]. Three fatal TTIs have emerged in the last 30 years: HIV infection, infection with West Nile virus (WNV), and infection with vCJD (variant Creutzfeldt-Jakob disease) prions. What happened in the past can happen again in the future and, if the next agent to emerge were concentrated in young
THE CASE FOR AN ALL-APHERESIS PLATELET SUPPLY

Figure 1. Relative risk (RR) of transmission of HIV, HCV, and HBV infection by transfusion of platelet pools versus single-donor (apheresis) platelets when a platelet pool tantamount to one adult therapeutic platelet dose consists of 4 WBD platelet concentrates (as assumed by the investigators of the Paul-Ehrlich Institute [1]) or of 5 or 6 WBD platelet concentrates (as is the case in the US and many European countries). All depicted differences in risk between pooled and single-donor platelet concentrates are statistically significant. The German investigators [1] adjusted the estimate of the incidence of HIV, HCV, and HBV infection in repeat donors for differences in the risk of infection by age group, gender, and population size of place of residence; and also for differences between whole-blood and plateletapheresis donors in age, gender, and population size of place of residence. (For example, 55% of whole-blood donors versus 73% of plateletapheresis donors were male, while males had a 7-fold higher risk of HIV infection, and a 3.6-fold higher risk of HBV infection than females.) After making all these adjustments, the investigators found that the RR of transmission in recipients of pooled versus single-donor platelets was 2.2 (95% CI, 2.1-2.4) for HIV, 2.7 (95% CI, 2.5-3.0) for HCV, and 3.2 (95% CI, 2.8-3.7) for HBV [1]. Here, the figure applies the same adjustments made by the German investigators (for age, gender, and population size of place of residence) to pools of at least 5 WBD platelet concentrates. Pools of at least 5 WBD platelet concentrates are generally necessary for meeting applicable standards for the minimal therapeutic platelet dose, so that—in policy debates—the RR of transmission in recipients of pooled versus single-donor platelets should be considered to be 2.75 for HIV, 3.375 for HCV, and 4.0 for HBV.

males from large urban areas in the manner that HIV was concentrated in young males from large urban areas, a move toward an all-apheresis platelet supply would not lessen the risk of transmission of the next major pathogen through platelet transfusion. The next major TTI to emerge is the cardinal threat to transfusion safety today, because a novel “HIV-like” pathogen could cause more deaths than TRALI (transfusion-related acute lung injury), TAS (transfusion-associated sepsis secondary to transfusion of a blood component contaminated with bacteria), and hemolytic transfusion reactions combined [3]. A novel “HIV-like” pathogen need not be a virus, however, and it need not be transmitted sexually. All it need have to reproduce the HIV catastrophe of the 1980s is a long asymptomatic phase during which donors will be making infectious donations.

Concerning the attribute of a chronic infection with prolonged pathogenemia, the closest to an “HIV-like” pathogen to emerge in the last 30 years have been the vCJD prions. It is also possible that the next major transfusion-transmitted pathogen could be “WNV-like”, that is, it could cause acute infection associated with a short and self-limiting period of pathogenemia. The micro-

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biologic characteristics of the next agent are totally unpredictable, as demonstrated by the emergence of the vCJD prions. Equally unpredictable is the agent’s transmission mode, because experience from the last 30 years has demonstrated that the agent can be transmitted sexually (like HIV), or it can be foodborne (like vCJD), or it can be vector-borne (like WNV), or it can even be airborne (like the coronavirus of the Severe Acute Respiratory Distress Syndrome [SARS]) [8-10].

Therefore, unlike the “traditional” transfusion-transmitted viruses (HIV, HCV, and HBV), which have well-described associations with particular age groups, male versus female gender, and residence in large cities and metropolitan areas versus small cities and rural areas, the next major transfusion-transmitted pathogen to emerge cannot be expected to be associated with any particular age group, or with male (versus female) gender, or with residence in large cities and metropolitan areas (versus small cities and rural areas). Based on the experience accumulated over the last 30 years, policy-makers must take a neutral position and assume that all blood donors could be equally at risk of contracting and transmitting the next pathogen, because neither the microbiologic characteristics nor the transmission mode of that agent can be predicted. Importantly, policy-makers should take into account the possibility that the accumulation of the next agent in the blood-donor population may have already commenced.

Thus, when it comes to modeling the relative risk (RR) of transmission of the next major transfusion-transmitted pathogen to emerge in the future by pooled WBD versus single-donor platelets, the adjustments made by the German investigators [1] (and which were most appropriate for modeling the residual risk of HIV, HCV, and HBV) must be removed from the model [1], because the epidemiologic of the next pathogen is unknown. If the next pathogen turns out to be “HIV-like” in the sense of being overly concentrated in young males residing in large cities and metropolitan areas, the German investigators [1] correctly predict that the over-representation of young males from large cities and metropolitan areas among plateletapheresis (versus whole-blood) donors could wipe out the difference in risk between pooled WBD versus single-donor platelets (so that platelet pools and apheresis platelets could end up having the same risk of transmitting the novel pathogen to transfusion recipients).

There is no reason, however, to a priori consider that the next pathogen will be “HIV-like” in the sense of being overly concentrated in young males residing in large urban areas. As already discussed, the next pathogen could be “HIV-like” in the same way that the vCJD prions are “HIV-like”: that is, associated with a long asymptomatic phase while being foodborne (rather than sexually transmitted). Alternatively, the next agent could be vector-borne or even airborne, showing little or no predilection for (or conceivably even showing aversion towards) young males residing in large urban areas. Accordingly, when it comes to modeling the RR of transmission of the next major transfusion-transmitted pathogen by pooled WBD versus single-donor platelets, the only determinant of the RR (before the epidemiologic characteristics of the next pathogen become known) is the number of donor exposures, that is, the number of WBD concentrates that make up a therapeutic platelet dose. Although the precise calculation of the RR is complex and involves conditional probabilities, the number of WBD concentrates in the pool makes for a very good approximation of the actual RR [2,3]. If the pools consist of 5 WBD concentrates (as is the case, on average, in the US [4]), policy-makers should assume that platelet pools have a 5 times higher risk of transmission of the next major transfusion-transmitted pathogen compared with single-donor (apheresis) platelets (Table 1 and Figure 2). If the pools consist of 4 WBD concentrates (as the German investigators have considered [1]), policy-makers should assume that platelet pools have a 4 times higher risk of transmission of the next agent compared with single-donor (apheresis) platelets. When the next agent actually emerges and its epidemiologic characteristics become known, the RR calculation should be adjusted based on the agent’s actual attributes, as was done for HIV, HCV, and HBV by the German investigators [1]. Such adjustments, however, ought to be corrections to the RR model made after the epidemiologic facts pertaining to the next agent become known; not assumptions about what these epidemiologic facts are bound to be.

In summary, based on the mathematical model presented by the Robert Koch Institute and the Paul-Ehrlich Institute [1], we now know that the RR of disease transmission by pooled WBD versus single-donor platelets is 2.2 (or 2.75 for pools of 5) for HIV, 2.7 (or 3.375 for pools of 5) for HCV, and 3.2 (or 4.0 for pools of 5) for HBV (Figure 1). This is the actual RR of transmission, by pools versus single-donor concentrates, of these “traditional” transfusion-transmitted viruses in Germany today. Beyond this actual risk, there is the theoretical risk of the next major transfusion-transmitted pathogen to emerge in the future. Although this risk is theoretical, it could cause a major catastrophe should it materialize, potentially resulting in more deaths than TRALI, TAS, and hemolytic transfusion reactions combined when transfusions of all blood components are considered, and even in a number of deaths comparable to half the total number of annual deaths from TRALI, TAS, and hemolytic transfusion reactions combined when transfusions of platelet pools alone are considered (Figure 2) [3]. If (or when) such a pathogen emerges, we know from the model of the German investigators [1] that the RR could be as low as 1.0 (if the next major transfusion-transmitted pathogen were “HIV-like” in the sense of being overly concentrated in young males residing in large urban areas), in which case the dire predictions of Figure 2 would not materialize. We also know, however, that the RR could be as high as 5.0 (for pools of 5) or 4.0 (for pools of 4) if no a priori assum-
Figure 2. Increase in the number of transfusion-transmitted infections with HIV, HCV, and HBV, as well as the next major “HIV-like” or “WNV-like” pathogen to emerge in the future if the 260,000 platelet doses still transfused as pools in the US in 2008 [4] are not replaced by single-donor (apheresis) platelets [2,3]. The increase in risk secondary to the continued use of platelet pools is calculated for the year preceding the introduction of blood-donor screening in the case of the next “HIV-like” pathogen; and for the duration of the first major seasonal epidemic of the next “WNV-like” pathogen. The “HIV-like” pathogen is an agent causing chronic infection with a long asymptomatic phase and prolonged pathogenemia during which blood donors are making infectious donations. Because of the surveillance systems that we now have, a novel “HIV-like” pathogen should not go undetected for as long as HIV went undetected. Nonetheless, it could attain a prevalence 10 times lower than the prevalence that HIV reached in US blood donors in 1984 (the year before blood-donor screening for HIV was introduced in 1985)—that is, the new pathogen could attain a prevalence of 1 per 10,000 donors [2,3,6,7] as the German investigators have assumed in the Paul-Ehrlich Institute model [1]. The “WNV-like” pathogen is an agent causing acute infection (with a short and self-limiting period of pathogenemia) in the course of a seasonal epidemic, as was the case with the first major epidemic of WNV in North America in the late summer and fall of 2002 (which resulted in 380 transfusion transmissions of WNV infection [11]). On average, a therapeutic platelet dose made up of pooled platelets in the US consists of 5 WBD platelet concentrates [4]. Thus, in 2008, the continued transfusion of 12.9% of all administered platelet doses as pooled WBD (rather than apheresis) platelets [4] resulted in 1.04 million preventable allogeneic-donor exposures [2,3]. These preventable exposures would result in 104 transmissions of the next major “HIV-like” pathogen to emerge if the agent attained a prevalence of 1 per 10,000 donors in the year before blood-donor screening was introduced. Although fewer than half of these recipients might survive long enough to develop disease [12], some 30 deaths could result for the sole reason that the 12.9% of therapeutic platelet doses now transfused as pools in the US were not replaced by single-donor platelets at the appropriate time. This number of deaths (about 30 deaths) is equal to the number of transfusion-related deaths from TRALI, TAS, and hemolytic transfusion reactions combined that occurred annually in the US between 2005 and 2010 (about 60 deaths) [2,3,5]. Initially published by Vamvakas in Transfusion 2012 (Epub ahead of print, Aug 6) [3].

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to transfusion of pooled rather than single-donor platelets. Although this difference in risk had long been known [14-17], it had not been previously documented by means of a mathematical model built by the incidence/window-period method. A longer-term action could be the prohibition of transfusion of pooled WBD platelets in Germany and the replacement of the platelet pools currently transfused by an all-apheresis platelet supply.

The purpose of this article is to examine the validity of possible arguments against such policy interventions. The arguments that will be considered here are that: 1) the available surveillance data from Germany do not confirm the increased risk of HIV, HCV, and HBV transmission from pooled WBD platelets compared with apheresis platelets predicted by the RKI/PEI model; 2) the magnitude of the difference in risk does not justify a policy intervention(s); and 3) the impending implementation of pathogen reduction (PR) of platelets will eliminate any difference in the risk of transmission of both known transfusion-transmitted viruses and emerging transfusion-transmitted pathogens between pooled WBD and single-donor (apheresis) platelets.

**Surveillance data do not confirm the increased risk of HIV, HCV, and HBV transmission from pooled WBD (compared with apheresis) platelets**

Between 1997 and 2010, 7 cases of HIV, HCV, or HBV infection acquired through platelet transfusion were reported to the Paul-Ehrlich Institute [18-23]. There were 0 HIV, 3 HCV, and 4 HBV infections. Of these, 2 infections (1 HCV and 1 HBV) were acquired from transfusion of pooled platelets, while 5 infections (2 HCV and 3 HBV) were acquired from transfusion of apheresis platelets [19,20]. Since 3,382,101 apheresis platelets and 2,280,338 platelet pools had been transfused between 1997 and 2010, the combined risk of HIV, HCV, and HBV transmission through platelet transfusion calculated from the German surveillance data was 1 per 676,000 apheresis platelets versus 1 per 1,110,000 platelet pools [18-23]. In this way, the German surveillance data indicated a 1.6-fold increase in the risk of HIV, HCV, and HBV transmission secondary to apheresis versus pooled WBD platelets [18-23]—which is the opposite from what would have been predicted by the RKI/PEI model [1].

In the case of HIV-, HCV-, and HBV-infected blood donors, infectivity in blood or plasma (number of infectious copies/milliliter) is generally so high that any patient transfused with even a few milliliters of blood or plasma generally contracts the infection. It is thus biologically implausible to postulate that, despite a 4-fold or greater increase in the number of donor exposures in association with platelet pools compared with single-donor platelets, platelet pools might have a lower risk of HIV, HCV, and HBV transmission compared with apheresis platelets, because the volume of plasma that enters the pool from the (single) infected donor contributing to the pool is several times smaller than the volume of plasma contained in an apheresis platelet component collected from an infected donor. It is equally implausible to postulate that the “dilution” of the infected donor’s 50 milliliters of plasma in a pool of 4 or more donors contributing equal amounts of plasma might reduce infectivity and prevent infection when only a few milliliters of infectious plasma generally suffice to transmit infection.

The reason for the discrepancy between what the RKI/PEI model [1] predicts and what the surveillance data [19-23] indicate should instead be sought in the well-documented difficulties inherent in interpreting any surveillance data. Adverse events captured by hemovigilance systems depend on whether a particular adverse event caused by a transfusion was suspected of being, investigated as possibly being, and/or deemed to be transfusion-related on the basis of the criteria employed locally for the diagnosis of each transfusion complication. Whether transfusion is considered as the possible cause of an adverse event varies with the medical and nursing staff awareness of transfusion complications at each particular clinical setting, as well as with local culture, resources, and logistics vis-à-vis the extent of investigation and the reporting of such adverse events along the designated channels of the hemovigilance system.

Thus, hemovigilance systems tend to greatly underestimate the incidence of transfusion-related adverse events. When TRALI was identified by a passive surveillance system relying on the Canadian Consensus Criteria [24] for making the diagnosis, the frequency of TRALI cases was in the range of 1 per 16,000 units of transfused plasma, 1 per 43,000 units of transfused platelets and 1 per 44,000 units of transfused red blood cells [25]. When TRALI was identified by a prospective observational study also employing the Canadian Consensus Criteria [24] for making the diagnosis in an intensive-care unit, 1 (8.2%) of 12 critically ill patients was reported to develop TRALI [26]. In the latter case [26], patients had generally received multiple blood components (red blood cells, plasma, and/or platelets) within 6 hours of developing acute lung injury (ALI) which met the definition of TRALI. [24]. Critically-ill patients (with sepsis, trauma, and other factors that may represent the “first hit” in the “two-hit” hypothesis of TRALI pathogenesis [27]) may have a lower threshold for TRALI than does the average transfusion recipient, so that they are more prone than average to develop TRALI. Some cases of ALI occurring in close proximity to a transfusion were also probably misdiagnosed as TRALI in this prospective observational study [26].

The > 1,000-fold difference in the reported incidence of TRALI between the passive surveillance [25] and the prospective observational study [26] is therefore partly due to the patient factors. It is undoubtedly also due, however, to the difference in study design (passive surveillance versus observational) and the tendency of the passive surveillance approach to greatly underestimate the incidence of transfusion-related adverse events.
When awareness of transfusion complications increases, or when resources are expended to detect transfusion complications, the reporting of transfusion complications to hemovigilance systems increases. Both of these conditions were satisfied, before the implementation of bacterial detection in platelets, at the Johns Hopkins Hospital (Baltimore, Maryland, US). Over 12 years, Ness et al. [14] had implemented a system of prospective monitoring, whereby all febrile reactions to platelets were assessed by culture. Under these conditions, the investigators observed a TAS risk of 1 per 3,000 platelet pools versus 1 per 15,000 apheresis platelets [14]. A virtually identical risk (1 per 2,282 platelet pools in 2000 and 1 per 4,149 platelet pools in 2001) was observed during the first 2 years of the Quebec hemovigilance program, which relied on hospitals that cultured 40.4% to 51.6% of platelet pools implicated in transfusion reactions and that were staffed with transfusion officers [28]. In contrast, from 1994 to 1998, the French hemovigilance system—probably the most comprehensive passive surveillance system that entailed mandatory reporting—observed a risk of TAS secondary to platelet transfusion of only 1 per 13,000 platelet pools [29]. The reported risk was far lower in the UK hemovigilance program [30,31].

Thus, surveillance systems grossly underestimate the risk of even acute adverse events following transfusion, such as TRALI or TAS. Surveillance systems are not designed to detect adverse events that do not occur in close proximity to the transfusion, such as transfusion-transmitted infections that do not cause clinical signs or symptoms until months or years after the transfusion. Such is the case with HIV, HCV, and HBV infection, of which several cases are expected to occur every year in the US based on the findings of the current incidence/window-period model [32], but hardly any are reported. Even after the implementation of testing for HIV and HCV RNA, at least 10 transmissions of HIV and 10 transmissions of HCV should occur annually in the US (given a risk of transmission for HIV of 1 per 2,135 million donations and a risk of transmission for HCV of 1 per 1,935 million donations [32]). Yet, only 11 (3.6%) of 307 transfusion-related deaths reported to the US Food and Drug Administration (FDA) from 2005 through 2010 had been due to a TTI other than TAS [5]—with none of these 11 deaths being due to HIV, HCV, or HBV infection.

In fact, surveillance systems are likely to miss even acute transfusion-transmitted infections, as has been demonstrated for TAS [33], and as was likely the case with some WNV infections occurring in New York in the summer of 1999 (when no transfusion-transmitted cases were reported in the course of the first WNV epidemic in North America), as well as with some dengue fever virus (DFV) infections occurring in endemic areas over many decades. The effectiveness of the current surveillance systems may thus be low, particularly at the point of recognition of events by physicians and their subsequent reporting to transfusion services [34]. As a result, the transmission of DFV through transfusion had gone undetected until recently [35,36]. The recording by the UK SHOT system of deaths from transfusion-acquired vCJD is due to the active surveillance efforts made in the UK to identify cases of transmission of vCJD through transfusion.

The reporting of 5 and 2 HIV, HCV, or HBV infections [19,20] attributed, respectively, to apheresis versus pooled platelets in 1997-2010 in Germany followed a considerable public debate that had taken place in 2011 [37] about the relative safety of pooled versus apheresis platelet components. The official German hemovigilance data for 1997-2010 [21-23] instead reported 6 (0 HIV, 2 HCV, and 4 HBV) infections from apheresis platelets and 1 (0 HIV, 1 HCV, and 0 HBV) infection from platelet pools (see Table 7 on page 26 of the 2010 hemovigilance report [21]). More specifically, the hemovigilance data for 2009 [22] (published on 3/30/2011; Table 7 [22]) and for 1997-2008 [23] (published on 4/27/2010; Table 8 [23]) had made no differentiation between pooled versus apheresis platelets vis-à-vis the number of TTIs attributed to each platelet component; only the latest (2010 [21]) hemovigilance report presented separate data on the number of TTIs reported to have been transmitted by pooled versus apheresis platelets. Even this latest [21] report published on 6/21/2012, however, did not present separate data on the number of cases of TRALI attributed to pooled versus apheresis platelets (see Table 3 on page 24). The change in the number of HIV, HCV, and HBV transmissions (5 versus 2 as opposed to 6 versus 1) ascribed to apheresis versus pooled platelets after the 2011 public controversy [37] reflected the uncertainty (owing to the limitations of any surveillance system in capturing precise information) about the type of platelet component from which the TTIs attributed to platelet transfusion had arisen.

In Germany, there is also a systematic difference between the clinical settings in which different platelet components are transfused. Platelet pools are primarily distributed by the Red Cross Blood Services to community hospitals. Apheresis platelets are primarily collected by university-hospital blood banks (or other hospital blood banks operated by large hospitals in large cities) and are transfused locally. If community hospitals transfuse platelets predominantly to patients with acute bleeding who are less likely to survive the transfusion (or to return for follow-up) than patients receiving platelets in the hematology and surgery units of university hospitals, we would expect many fewer patients from community hospitals (who receive platelet pools) than patients from university hospitals (who receive apheresis platelets) to be detected by the surveillance systems with transfusion-acquired HIV, HCV, or HBV infection. This could be because fewer of the former (than the latter) patients survive the transfusion or return for follow-up, or because (compared with the university-hospital setting) a transfusion-transmitted chronic infection is less likely to be diagnosed in the community.
hospital setting as being transfusion-acquired, and/or to be reported along the channels of the surveillance system.

Review of the overall versus the platelet pool-specific German surveillance data in 1997-2010 [21-23] lends credence to this hypothesis. On average over this period, there were about 4 to 4.5 million whole-blood donations annually in Germany. Approximately 20% to 25% of these whole-blood donations (or some 1 million donations annually) were used to produce the 220,000 platelet doses provided as pools of 4 or 5 WBD concentrates. Over 14 years (1997-2010), approximately 56 to 58 million whole-blood donations engendered 16 HBV transmissions with packed red blood cells that were detected and reported along the channels of the surveillance system. Up to 4 HBV transmissions would be likely expected to occur from platelet pools (if 20% to 25% of all whole-blood donations were used for making pools), but no HBV transmission was detected and reported [21]. Similarly, since 8 HCV transmissions were reported from 56 to 58 million donations of packed red blood cells in 1997-2010, 2 HCV transmissions would be expected from platelet pools, but only 1 was detected and reported [21]. Although these differences could certainly have occurred by chance, they may also reflect the fact that platelet pools are transfused specifically at community hospitals, while the other components made from whole-blood donations are transfused at either university or community hospitals.

Finally, donors of apheresis platelets are monitored intensively and over a long time for development of HIV, HCV, and HBV infection. Should an incident TTI be detected in an apheresis donor, lookback will be promptly initiated and potentially identify a case(s) of transfusion-transmitted infection that will be recorded by the surveillance system. This is less likely to happen with whole-blood donors who do not donate as frequently (or may not return to donate or may not notify the blood center of their diagnosis if they are diagnosed with HIV, HCV, or HBV infection in the community).

The reduction in risk is not of a magnitude sufficient to justify a policy intervention(s)

The incidence/window-period model (as used by the German investigators [1]) assumes that transfusion-transmitted viral infections arise because of donations made during the preseroconversion window period, considering that infections secondary to "immunosilence" infections in donors, infections secondary to variant viral strains, and infections secondary to laboratory errors (which are not taken into account by the model) are exceedingly rare [38]. In the early 1990s, during the infectious window period a donor was viremic, but did not yet have detectable HIV-1/2 antibody, HCV antibody, or HBsAg (hepatitis B surface antigen). When these infectious disease markers were used to detect incident infection, the mean length of the infectious window period was 22 days for HIV, 82 days for HCV, and 59 days for HBV [38]. The length of the window period could be reduced by the implementation of an additional test(s) for each transfusion-transmitted virus [38], and a considerable policy debate arose in the mid-1990s as to whether the reduction in risk made possible by the introduction of each additional screening assay was of a magnitude sufficient to justify the policy intervention. Table 2 shows the calculation of the RR of transmission of each transfusion-transmitted virus as a comparison of the risk of disease transmission in the absence of implementation of the proposed additional screening assay (respectively, HIV-1 p24 antigen or HIV-1 RNA, HCV RNA, or HBV DNA) versus the risk following the introduction of the additional screening assay. Based on a RR of 1.375 (and projected yield of 7) if testing for HIV-1 p24 antigen was not introduced in the mid-1990s, and a RR of 2.0 (and projected yield of 12) if testing for HIV RNA was not introduced in the late 1990s, US policy-makers proceeded to implement testing for HIV-1 p24 antigen in the mid-1990s, and then to replace the test for HIV-1 p24 antigen with a nucleic-acid technology (NAT) test for HIV-1 RNA in the late 1990s. Prior to introducing NAT testing for HIV-1, US policy-makers introduced NAT testing for HCV, because the anticipated benefit from testing for HCV RNA (RR of 3.565 and projected yield of 84—Table 2) was deemed to be greater than the anticipated benefit from testing for HIV RNA (RR of 2.0 and projected yield of 12). Although many US blood suppliers are testing blood donors for HBV DNA, the US FDA did not mandate that all US blood donors be screened for HBV DNA.

Thus, blood components screened only for anti-HIV-1/2 (or even anti-HIV-1/2 plus HIV-1 p24 antigen) are less safe than blood components screened for both anti-HIV-1/2 and HIV-1 RNA. This is not reflected merely in the labeling of the components (as a warning to transfusing clinicians), but—more importantly—in the prohibition of the use of components screened only for anti-HIV-1/2 (or anti-HIV-1/2 plus HIV-1 p24 antigen) and not screened for HIV-1 RNA as well. Similarly, blood components screened only for anti-HCV are less safe than blood components screened for both anti-HCV and HCV RNA. This is not reflected merely in the labeling of the components, but in the prohibition of the use of components screened only for anti-HCV (and not screened for HCV RNA as well).

Based on the precedent of these policy decisions made in the 1990s, and in the absence of a consideration of impending implementation of PR (discussed in the next section), pooled platelets must be considered less safe than apheresis platelets, because they have a 2.2-fold higher risk for pools of 4 (or a 2.75-fold higher risk for pools of 5) of transmitting HIV infection to transfusion recipients; and a 2.7-fold higher risk for pools of 4 (or a 3.375-fold higher risk for pools of 5) of transmitting HCV infection to transfusion recipients. The RR for pooled versus apheresis platelets in the case of HIV (2.2 or 2.75) exceeds the RR that led to the introduction of screening for HIV-1 p24 antigen (1.375) or HIV-1 RNA.
Table 2. Relative risk of transmission of HIV, HCV, or HBV infection by transfusion: risk in the absence of implementation of the proposed additional screening assay versus risk following the introduction of the additional test*.

<table>
<thead>
<tr>
<th>Transfusion-transmitted virus</th>
<th>Infectious disease marker representing incident infection</th>
<th>Estimate of the mean length of the infectious window period (days)</th>
<th>Projected yield from the reduction in the length of the window period thanks to the introduction of a new (additional) screening assay†</th>
<th>Relative risk of the residual risk of transmission of the corresponding viral infection (risk in the absence of implementation of the additional test versus risk following the introduction of the additional screening assay)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV</td>
<td>Anti-HIV-1/2</td>
<td>22</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>HIV-1 p24 antigen</td>
<td>16</td>
<td>7</td>
<td>1.375 (reduction of the window period by 6 days and starting window period of 22 days [a])</td>
</tr>
<tr>
<td></td>
<td>HIV-1 RNA</td>
<td>11</td>
<td>12</td>
<td>2.0 (reduction of the window period by 11 days and starting window period of 22 days [b])</td>
</tr>
<tr>
<td>HCV</td>
<td>Anti-HCV</td>
<td>82</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>HCV RNA</td>
<td>23</td>
<td>84</td>
<td>3.565 (reduction of the window period by 59 days and starting window period of 82 days [c])</td>
</tr>
<tr>
<td>HBV</td>
<td>HBsAg</td>
<td>59</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td></td>
<td>HBV DNA</td>
<td>34</td>
<td>81</td>
<td>1.735 (reduction of the window period by 25 days and starting window period of 59 days [d])</td>
</tr>
</tbody>
</table>

* Based on data reported by Schreiber et al [38] in 1996 and reflecting the capacity of tests available at that time to detect infectious disease markers of HIV, HCV, or HBV infection. For example, the window period prior to the appearance of HCV antibody refers to HCV antibody detected by second generation enzyme immunoassays (HCV EIA 2.0). This was the evidence based on which the policy decisions were made in the 1990s to introduce mandatory testing for HIV-1 RNA and HCV RNA.

† That is, absolute number of infectious donations intercepted annually in the US thanks to the introduction of the corresponding new (additional) test for each transfusion-transmitted virus, as reported by Schreiber et al [38]. The projected yield estimates are based on the findings of the incidence/window-period model [38] and not on surveillance data. They were not reproduced (nor were they expected at the time to be reproduced) by empirical evidence of actual transfusion-transmitted infections. Even so, the number of infectious donations intercepted annually is high in the Table compared with current perception, because the risk of transmission of each virus at the time [38] was significantly higher than it is today [32]: 1 per 493,000 (versus 1 per 2,135 million donations today) for HIV; 1 per 103,000 (versus 1 per 1,935 million today) for HCV; and 1 per 63,000 (versus 1 per 280,000 today) for HBV. Because Germany is 4 times smaller than the US, the relevant projected-yield figures would be about 3 (rather than 12) for HIV-1 RNA and 21 (rather than 84) for HCV RNA. These figures might have been even lower for Germany in the 1990s if—as is the case today [32,39]—the risk of HIV and HCV transmission in the 1990s was lower in Germany compared with the US.

a That is, 22 divided by 16 equals 1.375
b That is, 22 divided by 11 equals 2.0
c That is, 82 divided by 23 equals 3.565
d That is, 59 divided by 34 equals 1.735
N/A = Not applicable.
than 4-fold) with pools of 4 compared with apheresis respectively [39]; and 3) the risk of HIV, HCV, and HBV million, 1 per 10.9 million, and 1 per 360,000 units, respectively [39]; and 3) the risk of HIV, HCV, and HBV transmission is increased 2.2-, 2.7-, or 3.2-fold (rather than 4-fold) with pools of 4 compared with apheresis platelets [1].

The absolute number of 1 to 2 HIV-, HCV-, or HBV-infectious platelet doses intercepted annually in Germany is much smaller than the projected yield (Table 2) from the implementation of screening for HIV-1 RNA and HCV RNA in the 1990s. The replacement of platelet pools by an all-apheresis platelet supply, however, becomes proportional (in terms of yield) to the introduction of screening for HIV-1 RNA and HCV RNA in the 1990s when we also consider the next major transfusion-transmitted pathogen to emerge in the future. If this pathogen is “HIV-like” in the sense of having a prolonged period of asymptomatic pathogenemia during which donors will be making infectious donations, and if it attains a prevalence of 1 per 10,000 German donors during the year preceding the timely implementation of blood-donor screening for the new pathogen, the implementation of an all-apheresis platelet supply is thus not associated with any risk to donors (Figure 3) [45].

In contrast to what the historical literature on donor reactions reviewed by Schrezenmeier and Seifried [46] has indicated, this modern approach to meeting all of a country’s platelet transfusion needs through apheresis should not be associated with any increase in moderate and severe donor reactions in Germany, as it has not been associated with any increase in moderate and severe donor reactions in the US (Figure 3) [45]. On the contrary, the rate of moderate and severe donor reactions should decrease with an all-apheresis platelet supply (as has been the case in the US [45]) if plateletapheresis is performed with the cell separators used today and at the hands of trained personnel who have accumulated significant experience with the performance of the procedure. Equally importantly, if an all-apheresis platelet supply becomes part of the patient-centric paradigm of clinical transfusion practice for the 21st century
Figure 3. Odds ratio (OR) of moderate or severe donor reactions with platelet/plasmapheresis or "double" red-blood-cell collection, or red-blood-cell collection with concomitant collection of plasma and/or platelets, compared with whole-blood collection [45]. These data are based on 793,293 allogeneic collections (including 552,183 whole-blood, 54,841 platelet/plasmapheresis, and 164,179 “double” red-blood-cell collections, as well as 18,790 red-blood-cell collections combined with collections of plasma and/or platelets) made at the blood centers of one US blood provider (Blood Systems) in 2007; and on a multivariate model into which (in addition to collection type) the following variables were eligible for entry: donation site (fixed versus mobile), association type (high school, college, or other), age, gender, race, donation history, blood volume, weight, body mass index, pulse, systolic blood pressure, diastolic blood pressure, and blood center identifier [45]. Each OR is surrounded by its 95% CI. Provided that the 95% CI of the OR does not include the null value of 1, an OR < 1 indicates a lower (p < 0.05) donor reaction rate with each of the three types of apheresis collections compared with whole-blood donation. When plateletapheresis is performed with the cell separators available today—and at the hands of trained personnel who have accumulated significant experience with the performance of the procedure—donors who self-select for a plateletapheresis collection have a lower (p < 0.05) reaction rate compared with whole-blood donors. Based on data reported by Kamel et al. [45] and initially published by Vamvakas in Vamvakas EC: Decision-Making in Transfusion Medicine; Bethesda, MD: AABB Press, 2011, p. 134.
trade-off as do policy-makers in the environmental sector: that is, a trade-off between the economic cost of introducing a precautionary measure and the potential health benefits that the proposed measure might provide [48,49]. The precautionary principle—enshrined in European Union law [50]—states that, when a purported risk represents a threat of “serious or irreversible damage” (as a fatal transfusion-acquired disease would do), complete evidence of risk does not have to exist to justify the institution of measures to protect individuals and society from that risk. If a new pathogen with “unpredictable” characteristics (along the lines of the vCJD prions) were to emerge in the future, and/or if PR of platelets were not ripe for implementation (as considered in the next section), there is no alternative to the reduction in the number of donor exposures (that an all-apheresis platelet supply would confer) for containing an epidemic of platelet transfusion-acquired disease.

Mutations are frequent in RNA viruses (such as HIV and HCV), and when mutations result in aminoacid substitutions, the antigenicity of the virus may be affected, and the antibody produced by the host may not cross-react adequately with the antigens used for capture in screening assays [51]. In addition to such false-negative antibody screening test results, HIV-1 RNA false-negative screening assay results are possible if a mutation occurs in the single RNA region targeted by a (single-target) RNA screening assay [52].

Currently in Germany, HIV-1 RNA tests that amplify a single target region of the HIV-1 RNA genome, and that detect a minimum of 10,000 viral copies/milliliter, meet the PEI requirements for HIV-1 RNA screening of blood donors [52]. Between 2007 and 2010, the Paul-Ehrlich Institute asked German blood operators to perform investigations of samples suspected of producing false-negative HIV-1 RNA test results. Based on the responses received from the blood operators, 17 cases of false-negative HIV-1 RNA assay results (in donors with documented HIV infection) were observed between 2007 and 2010 [52]. In 14 cases, the HIV-1/2 antibody test results had been positive, and the donations were thus intercepted. In 3 cases, however, the HIV-1/2 antibody test results were negative, and the units were transfused, resulting in 2 documented cases of HIV transmission by transfusion [52,53]. Further investigation showed that the false-negative HIV-1 RNA assay results were due to HIV-1 viral strains in which the (single) HIV-1 RNA region targeted by the employed HIV-1 RNA test was mutated, so that this RNA region was either under-recognized or not recognized at all by the HIV-1 RNA assay [52,53].

Based on these active-surveillance data, the Paul-Ehrlich Institute estimated the risk of HIV transmission (secondary to mutations of the single RNA region targeted by the HIV-1 RNA tests) to be 1 per 9.64 million donations [52], or approximately 1 infectious donation made every 2 years. This risk of HIV transmission is comparable to the absolute increase in the risk from platelet pools for HIV, HCV, and HBV transmission combined (1 to 2 HIV-, HCV-, or HBV-infectious platelet doses annually).

Appropriately, the Paul-Ehrlich Institute intervened to interrupt this risk of HIV transmission by transfusion, and it determined that—within 30 months of the initial communication [52]—German blood operators should implement “dual-target” RNA screening assays (i.e., assays that target two separate regions of the HIV-1 RNA genome for amplification); or implement alternate RNA tests that afford recipients the same level of protection as the “dual-target” RNA screening tests do [52]. Given this precedent [52], the Paul-Ehrlich Institute should intervene in the same manner to intercept the 1 to 2 HIV, HCV-, or HBV-infectious platelet doses annually that are assumed to occur based on the findings of the RKI/PEI mathematical model [1]. Even if the implementation of “dual-target” HIV-1 RNA screening tests was deemed to be virtually cost-neutral [52], whereas the move toward an all-apheresis platelet supply is regarded as costly, policy-makers should consider that the move toward an all-apheresis platelet supply will also reduce the risk of transmission of the next major transfusion-transmitted pathogen(s) to emerge in the future; whereas no such benefit will be conferred by the implementation of “dual-target” HIV-1 RNA screening assays by January 1, 2015.

The impending implementation of pathogen reduction of platelets will eliminate any difference in the risk of transfusion-transmitted infections between platelet pools and single-donor platelets

If PR of platelets were ripe for implementation (i.e., demonstrated to be free of possible adverse effects introduced by the PR process, which perhaps equal or exceed the benefit derived from the prevention of infectious-disease transmission), PR of platelets could indeed eliminate the residual risk of transmission of HIV, HCV, and HBV infection, as well as prevent transfusion transmission of emerging pathogens containing nucleic acids. With the same proviso, PR of platelets could prevent the very important transfusion risk of TAS secondary to bacterial contamination of platelet components. PR could not protect from novel agents not containing nucleic acids (along the lines of the vCJD prions), but the extent of protection afforded by PR vis-à-vis the traditional (nucleic-acid-based) pathogens would be of a magnitude sufficient to recommend its implementation [6,7,54].

The recommendation for the use of PR [54], however, was made (and PR systems for platelets were approved in Europe as early as 2006) before the randomized controlled trial (RCT) of Kerkoffs et al. [55] reported a statistically significant (p < 0.05) increase in the risk of bleeding between recipients of pathogen-reduced (versus non-pathogen-reduced) platelets; and before a meta-analysis [56] of all available RCTs comparing the risk of bleeding between recipients of pathogen-reduced ver-
Figure 4. Mechanism postulated by Hitzler and Vamvakas [77], and by Osman, Hitzler, Hellstern, Vamvakas, and Provost (4th International StKB Future Workshop on Hema and Cell Therapy: Platelet microRNA profiles and the effect of pathogen reduction on platelet function; Mainz, Germany; April 20, 2012) to explain the increase in bleeding complications in association with PR observed by the RCT of Kerkhoffs et al. [55] and by the meta-analysis of all available RCTs [56,78] comparing the risk of bleeding in recipients of pathogen-reduced versus non-pathogen-reduced platelets. Investigation of this hypothesis will require direct comparisons of the levels of specific platelet microRNAs (selected from a list of microRNAs that are abundantly and consistently expressed in platelets), and of the function of pathogen-reduced versus untreated platelets, over 7 days of storage following PR.

The recommendation for the use of PR [54] was also made before platelets were known to contain significant amounts of functionally-important nucleic acids. We now know that platelets retain a diverse transcriptome from their megakaryocytic precursors [61], so that up to one-third of all human genes are present in platelet mRNAs [62-65]. Platelets make use of their mRNAs to template de novo protein synthesis both in the circulation and during platelet storage in the blood bank [66-72]. Platelets also contain an especially abundant and diverse array of microRNAs—that is, small, 19- to 24-nucleotide-long, non-coding RNA species—and indeed a functioning microRNA pathway that regulates mRNAs and gene transcription and may thus affect platelet protein translation and platelet function [71-76].

Given the increase in bleeding complications reported in recipients of pathogen-reduced (versus non-pathogen-reduced) platelets by Kerkhoffs et al. [55] and by the meta-analysis of all RCTs [56], PR might impair the function of all treated platelets in addition to causing the known loss of a proportion of the treated platelets [55,56]. More specifically, PR could reduce the platelet content of microRNAs by inhibiting microRNA synthesis through crosslinking of their double-stranded precursors (if PR targeted nucleic acids as short as these double-stranded precursors) [77]. Alternatively, the cellular perturbation caused by PR might cause release of microRNAs from platelets, either directly into the supernatant or inside microvesicles. Either way, there could be reduced microRNA content of the PR-treated platelets, impaired protein synthesis during their storage in the blood bank, and impairment of their function which could explain the increased bleeding observed in recipients of treated (versus untreated) platelets [55,56], despite the compensatory platelet transfusions given to...
Figure 5. Odds ratios (ORs) and associated 95% CIs calculated separately for all bleeding (WHO or CTC bleeding grades 1 through 4), clinically significant bleeding (WHO or CTC bleeding grades 2 through 4), and severe bleeding (categorized as such by the investigators or bleeding of WHO or CTC grades 3 and 4), across 5 [55,57-59,79], 2 [55,58], and 4 [55,57-59] RCTs that reported on each outcome in patients randomized to receive either pathogen-reduced with the Intercept system or untreated platelets [78]. Each OR is surrounded by its 95% CI. Provided that the 95% CI of the OR does not include the null value of 1, an OR > 1 indicates a greater (p < 0.05) risk of bleeding complications in recipients of pathogen-reduced versus non-pathogen-reduced platelets. Clinically-significant bleeding was significantly (p < 0.05) increased in association with PR across the SPRINT trial [58] and the RCT of Kerkhoffs et al. [55], but the increase in all bleeding and in severe bleeding did not attain significance. An increase in clinically-significant bleeding was observed (as shown in the figure) when the meta-analysis integrated the findings on bleeding complications from the SPRINT trial as presented in the expanded safety analysis [58], but not the initial report of that study [81]. For a comparison of the bleeding complications in the SPRINT trial as presented in the expanded safety analysis [58] versus the initial report of that study [81], see Vamvakas [78]. Initially published by Vamvakas in Vox Sang 2012;102:302-16 [78].

Figure 6. Odds ratios (ORs) and associated 95% CIs, for WHO grade 2 bleeding in the SPRINT trial [81]; all bleeding (WHO or CTC bleeding grades 1 through 4) in the euroSPRITE trial [57], the RCT of Janetzko et al. [59], and the RCT of Kerkhoffs et al. [55]; and WHO grade 2 bleeding in the RCT of Lozano et al. [79], as combined in the meta-analysis of Cid et al. [80]. Owing to clinical heterogeneity resulting from the attempt to combine such clinically heterogeneous outcome measures, there was moderate statistical heterogeneity (p = 0.08 for the Q test statistic) in this meta-analysis [80], which had not been present in the meta-analysis of the same studies by Vamvakas [78] because the latter meta-analysis had solely integrated results pertaining to clinically homogeneous outcomes (i.e., results on all bleeding or clinically-significant bleeding or severe bleeding). When Cid et al. [80] combined the study results on these clinically heterogeneous outcome measures (as shown in the above figure), they found—in the presence of the aforementioned moderate statistical heterogeneity—no difference in bleeding complications between recipients of pathogen-reduced versus non-pathogen-reduced platelets (summary OR = 1.13; 95% CI, 0.72-1.78). To resolve the problem of moderate statistical heterogeneity, Cid et al. [80] then excluded from the meta-analysis the RCT of Kerkhoffs et al. [55]. The RCT of Kerkhoffs et al. [55] had been the only study to find a statistically significant (p < 0.05) increase in the risk of bleeding complications in association with PR. It was also the only unblinded study, the only independently-funded (as opposed to manufacturer-sponsored) trial, and the only study to enroll an unselected patient population (i.e., to include patients with all risk factors predisposing to platelet consumption). After the RCT of Kerkhoffs et al. [55] was excluded, Cid et al. [80] calculated a summary OR of 0.97 (95% CI, 0.75-1.27), that is, they again found no difference in bleeding complications between recipients of pathogen-reduced versus non-pathogen-reduced platelets. The summary ORs calculated by Cid et al. [80] before and after the exclusion of the RCT of Kerkhoffs et al. [55] are not plotted in the above figure, owing to the inappropriateness of combining such clinically heterogeneous outcome measures (i.e., grade 2 bleeding in the SPRINT trial [57] and the meta-analysts’ own RCT [79] versus all bleeding in the 3 other RCTs [55,57,59]) to calculate summary ORs by a meta-analysis.
make up for the platelet losses known to be caused by PR (Figure 4).

Two meta-analyses [56,78] by Vamvakas (one including the single Mirasol [60] trial and one limited to the Intercept studies [55,57-59,79]), and one meta-analysis [80] of the Intercept RCTs [55,57-59,79,81] by Cid et al., have reached discrepant conclusions as to whether PR increases the risk of bleeding complications in settings in which patients receiving pathogen-reduced platelets also receive prophylactically compensatory platelet transfusions to make up for the platelet losses known to be caused by PR (and thus maintain a platelet count of at least 10,000/µL). The discrepancy in the conclusions reached by the two groups is remarkable, because the meta-analysis by Cid et al. [80] and the second meta-analysis by Vamvakas [78] integrated the results of the same 5 Intercept RCTs [55,57-59,79,81]. Vamvakas [78] separately integrated the reported results on all bleeding (World Health Organization [WHO] or National Cancer Institute Common Toxicity Criteria [CTC] bleeding grades 1 through 4), on clinically significant bleeding (WHO or CTC bleeding grades 2 through 4), and on severe bleeding (i.e., bleeding categorized as such by the authors of the RCT or bleeding of WHO or CTC grades 3 and 4). The meta-analysis thus integrated clinically homogeneous outcome measures and each of the three performed analyses was correspondingly statistically homogeneous. There was a statistically significant (p < 0.05) increase in clinically significant bleeding across the studies in association with PR, albeit not in all bleeding or severe bleeding (Figure 5). Concerning the definition of severe bleeding, briefly, grade 1 bleeding encompasses mild bleeding for which no intervention is needed; grade 2 bleeding encompasses symptomatic bleeding for which some intervention (although not transfusion) is needed; grade 3 bleeding requires transfusion or other intervention; and grade 4 bleeding is catastrophic.

Cid et al. [80] presented only a single integration of the bleeding complications presented in the published RCTs [55,57-59,79,81], in which they combined the results on grade 2 bleeding from the largest (SPRINT) trial [81] with the results on all bleeding from 3 other RCTs [55,57,59]. Thus, symptomatic bleeding for which some intervention (although not transfusion) was needed in the largest RCT [81] (which enrolled more than half of the total number of patients available for meta-analysis) was combined with all bleeding (i.e., mild bleeding for which no intervention is needed, symptomatic bleeding for which some intervention is needed, bleeding requiring transfusion or other intervention, as well as catastrophic bleeding) in 3 other studies [55,57,59]. This combination of clinically heterogeneous outcome measures (i.e., grade 2 bleeding in the SPRINT trial [81] versus grade 1 through grade 4 bleeding in 3 other RCTs [55,57-59]) suggested no difference in bleeding between recipients of pathogen-reduced versus non-pathogen-reduced platelets (Figure 6). However, whether such a combination of clinically heterogeneous outcome measures has any biologic meaning or clinical relevance is questionable. There have been two reports of the bleeding complications observed in the large SPRINT trial [58,81]. The initial report [81] (on which Cid et al. [80] relied) made daily, per-protocol assessments of bleeding by blinded assessors, on the WHO scale and during only the period of platelet transfusion support. The expanded safety analysis [58] (on which Vamvakas [78] relied) compiled spontaneous reports of bleeding by blinded on-site personnel, on the CTC scale and during both the period of platelet transfusion support and an ensuing surveillance period (i.e., a period of observation similar to that used in the other RCTs). The expanded safety analysis [58] reported separately on grade 1, grade 2, grade 3, and grade 4 bleeding. The initial report [81] reported only on grade 2, grade 3, and grade 4 bleeding, while it designated only grade 2 bleeding as the study’s primary outcome, leading Cid et al. [80] to integrate solely the SPRINT trial results on grade 2 bleeding in their meta-analysis. Although the initial report reflected the experimental design, the expanded safety analysis [58] was conducted because of concerns voiced about the analysis of adverse events as presented in the initial report [81]. An analysis of the differences in the frequency of bleeding complications presented in the two reports of the SPRINT trial [58,81] (included as Figure 1 in Vamvakas’ meta-analysis [78]) accounts for the difference in the conclusions reached by Vamvakas [78] and by Cid et al. [80].

Before PR can be considered ready for implementation, we must investigate whether PR reduces the level of platelet microRNAs, thereby potentially impairing platelet function and causing increased bleeding in recipients (Figure 4). Further RCTs are also needed to resolve this question [82], and an RCT of the Mirasol system is currently under way in the Netherlands and Canada. It is possible that these pending studies will show no effect of PR on the level of platelet microRNAs or no increase in bleeding or other adverse effects secondary to PR, establishing the safety of PR and making PR ready for implementation. Alternatively, these studies may show that the implementation of PR systems for platelets (which are based on chemically-induced cross-linking of nucleic acids) is inappropriate, because platelets contain significant amounts of functionally-important nucleic acids which are reduced by PR, leading to an impairment of platelet function in PR-treated platelets.

**DISCUSSION**

Recently, the Robert Koch Institute and the Paul-Ehrlich Institute presented the first mathematical model finding a statistically significant increase in the risk of HIV, HCV, and HBV infection by platelet transfusion when pools of 4 WBD concentrates, rather than 1 single-donor (apheresis) component, are used to provide
one adult therapeutic platelet dose [1]. The relative increase in the risk of transmission of HIV, HCV, and HBV infection secondary to receipt of a platelet pool rather than a single-donor platelet component (RR of 2.2 or 2.75 for HIV, 2.7 or 3.375 for HCV, and 3.2 or 4.0 for HBV, with pools of 4 or 5 WBD concentrates, respectively—Figure 1) is similar to the relative increase in risk secondary to receipt of a unit not screened for HIV-1 RNA and HCV RNA (in addition to the antibody screening tests) versus receipt of a unit fully screened by the tests required by regulators today. The absolute increase in the risk from pools (number of HIV, HCV, and HBV transmissions that could be prevented annually if the platelet pools were replaced by apheresis platelets) is much smaller than the projected yield (Table 2) from the implementation of screening for HIV-1 RNA and HCV RNA in the 1990s. The replacement of platelet pools by an all-apheresis platelet supply, however, becomes proportional in terms of yield to the introduction of screening for HIV-1 RNA and HCV RNA in the 1990s when we also consider the next major transfusion-transmitted pathogen to emerge in the future. Although the replacement of platelet pools by an all-apheresis platelet supply can be expected to intercept only 1 to 2 HIV-, HCV-, or HBV- infectious platelet doses annually, it could prevent up to 88 transmissions of the next “HIV-like” pathogen to emerge in the future during the year preceding the implementation of testing of the new pathogen. (Such would be the case if the new pathogen attained a prevalence of 1 per 10,000 donors and no a priori assumptions were made about the epidemiology and/or microbiologic characteristics of the new pathogen.)

Both the reduction in risk thanks to the implementation of additional screening assays debated in the 1990s and the reduction in risk thanks to the implementation of an all-apheresis platelet supply debated presently have been based on mathematical models using the incidence/window-period method. This is the only appropriate basis for making policy decisions concerning measures to reduce or prevent disease transmission by transfusion, because surveillance data grossly underestimate the incidence of even acute adverse transfusion events, and they are not designed to capture chronic infections contracted by transfusion and becoming manifest only months or years following the transfusion. When rare events (such as TTIs) are recorded by surveillance systems, there is certainty about the existence of risk, although the magnitude of the risk cannot be determined from the surveillance data. Risk-management action should be taken in the absence of certainty about risk (such as would be secured from surveillance data); mathematical models are necessary for quantifying the risk of rare events (such as TTIs) and for guiding the risk-management action (so that the risk-management action can be both proactive and proportionate to the magnitude of the risk) [83].

The projected yield shown in Table 2 (number of HIV- or HCV-infectious donations intercepted annually thanks to the implementation of HIV-1 RNA and HCV RNA screening) was not recorded by either US or German surveillance systems. Nonetheless, the figure derived from the incidence/window-period model [38] at that time (Table 2) was the best available estimate of the expected benefit from the introduction of HIV-1 RNA and HCV RNA screening, and these additional screening tests were implemented in both the US and Germany. Their introduction prevented (since the late 1990s) many cases of transfusion-acquired HIV or HCV infection that would not have been captured by our surveillance systems had they been permitted to occur.

Although implementation of PR of platelets would eliminate the difference in the risk of transmission of HIV, HCV, and HBV infection between platelet pools and single-donor platelets, the hitherto-developed PR systems for platelets are not ready for implementation. The recommendation for the use of these PR systems [54] (which are based on chemically-induced cross-linking of nucleic acids) was made before theRCT of Kerkhoffs et al. [55] found a statistically significant increase in the risk of bleeding between recipients of pathogen-reduced (versus non-pathogen-reduced) platelets; and before platelets were known to contain significant amounts of functionally-important nucleic acids. Currently, further studies are required to determine whether PR is associated with increased bleeding and/or other adverse effects [82]. Until the safety or PR is established, it is inappropriate to transfuse pathogen-reduced platelets outside the setting of an RCT.

The immediate benefit from PR would be the prevention of ≥ 90% cases of TAS (i.e., of all cases of TAS except for the very rare cases secondary to spore-forming bacteria). However, we now have an alternative solution for this major transfusion risk (despite the logistic difficulties involved): a second bacteria detection test performed at the time of issue of the component, which can prevent 70% to 90% of cases of TAS [84,85]. Nonetheless, PR can prevent both TAS and infection with the next major transfusion-transmitted pathogen to emerge in the future (if the next agent contains nucleic acids). Thus, if shown to be safe, PR should replace the bacteria detection tests to protect against both TAS and the next major TTI to emerge in the future. When the time for the implementation of PR comes, however, PR should be performed on an all-apheresis platelet supply (Table 3).

The time for converting to an all-apheresis supply is now, since: 1) an all-apheresis supply confers a benefit of sufficient magnitude to justify this policy decision based on the precedent of implementing HIV-1 RNA and HCV RNA screening in the 1990s (Table 2); and 2) an all-apheresis platelet supply is not associated with any risk to either patients or donors (Figure 3) [45] if donation frequency does not increase [42-44]. The time for introducing PR will be when (or if) the safety of PR is established. Importantly, the findings of the on-going or planned studies of the safety of PR cannot be predicted. Therefore, the implementation of an all-aphere-
Table 3. Three alternate risk-reduction strategies for mitigating the risk of emerging TTIs secondary to platelet transfusion†.

- **Transfusion of non-pathogen-reduced single-donor platelets if:**
  - the available PR systems result in unacceptable risks of bleeding complications; *and*
  - there is no platelet PR system capable of inactivating all transfusion-transmitted pathogens

- **Transfusion of pathogen-reduced pooled WBD platelets if:**
  - PR can be achieved with a tolerable increase in bleeding complications; *and*
  - the reduction in the number of allogeneic-donor exposures does not result in other (e.g., immunologic, such as TRALI prevention) benefits

- **Transfusion of pathogen-reduced single-donor platelets if:**
  - the reduction in the number of allogeneic-donor exposures results in other (e.g., immunologic, such as TRALI prevention) benefits; *or*
  - there is no PR system capable of inactivating all emerging pathogens *provided that*
  - PR can be achieved with a tolerable increase in bleeding complications

† Decision criteria pertaining to the situation after systems for PR of platelets become licensed by the US FDA. Currently, transfusion of pathogen-reduced platelets should not be considered outside the experimental setting of an RCT. Initially published by Vamvakas in Vamvakas EC: Decision-Making in Transfusion Medicine; Bethesda, MD: AABB Press, 2011, p. 291.

Table 4. Component-centric versus patient-centric philosophies for meeting patient needs for platelet transfusion.

<table>
<thead>
<tr>
<th>Component-centric</th>
<th>Patient-centric</th>
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<tbody>
<tr>
<td>Either relies primarily (or exclusively) on transfusion of pooled WBD platelets (for all patients except patients needing HLA-matched platelet support), or permits the continued transfusion of pooled WBD platelets for reasons other than inexorable supply needs or the limited resources of a country’s healthcare system†</td>
<td>Either prohibits the transfusion of pooled WBD platelets altogether, or limits the transfusion of pooled WBD platelets to the minimum imposed on policy-makers by inexorable supply needs or the limited resources of a country’s healthcare system†</td>
</tr>
<tr>
<td>Suspends pooled WBD platelets in 100 mL of plasma from a male donor contributing to the pool, to reduce the risk of “alloimmune” TRALI*</td>
<td>Limits transfused platelets to single-donor platelets collected from male donors or screened female donors, to eliminate the risk of “alloimmune” TRALI</td>
</tr>
<tr>
<td>Administers red blood cells and plasma from different allogeneic whole-blood donors to multitransfused platelet transfusion recipients needing transfusion of red blood cells and plasma in addition to platelets</td>
<td>Reserves the platelets and plasma (and/or red blood cells) harvested from the same multicomponent-apheresis collection for transfusion to the same recipient</td>
</tr>
<tr>
<td>Maintains a platelet count of at least 10,000/µL in patients with hypoproliferative thrombocytopenia by transfusing a pool of WBD platelets each time that a patient’s platelet count falls below 10,000/µL</td>
<td>Maintains a platelet count of at least 10,000/µL in patients with hypoproliferative thrombocytopenia by transfusing half an apheresis platelet concentrate each time that a patient’s platelet count falls below 10,000/µL; and by reserving the other half of that apheresis component for transfusion to the same recipient the next time that that patient needs prophylactic platelet transfusion</td>
</tr>
<tr>
<td>Will presumably pathogen-reduce pooled WBD (or both pooled WBD and single-donor) platelets when systems for PR of platelets are licensed by the US FDA, for reasons not limited to inexorable supply needs or the limited resources of a country’s healthcare system</td>
<td>Will pathogen-reduce only single-donor platelets (along with any minimum of pooled WBD platelets imposed on policy-makers by inexorable supply needs or the limited resources of a country’s healthcare system) when systems for PR of platelets are licensed by the US FDA</td>
</tr>
</tbody>
</table>

† Preventing the recruitment of an adequate number of plateletapheresis donors or the acquisition of donor apheresis technology.
When the patient-centric paradigm of the 21st century is implemented, low-dose recipients of apheresis concentrates can thus be supported through the period of hypoproliferative thrombocytopenia with a median of 2.5 donor exposures compared with 18 donor exposures for recipients of pools of 6 WBD concentrates (5 transfusion events in which a total of 2.5 apheresis concentrates are administered in the former case versus 3 transfusion events in which a total of 3 pools of 6 WBD concentrates each are administered in the latter case). When platelet dosing is optimized in this manner [92] and the two halves of each apheresis concentrate are transfused to the same recipient in accordance with the principles of the patient-centric paradigm of clinical transfusion practice (Table 4) [3,90], the ratio of donor exposures in recipients of pooled versus single-donor platelets during the period of platelet transfusion support for hypoproliferative thrombocytopenia is 18 to 2.5, or 7.2. The ratio of donor exposures (7.2) in this population of platelet transfusion recipients is thus considerably higher than the ratio of 4 (assumed by the German investigators') or the ratio of 5 (depicted in the middle section of Figure 1 and used in most calculations presented in this article). Thus, when we consider what an all-apheresis platelet supply can do for patients in the context of the patient-centric paradigm of the 21st century [3,87,90], both the estimate of the number of HIV, HCV, and HBV transmissions predicted by the RKI/PEI model (1 to 2 transmissions annually because platelet pools have not been replaced by an all-apheresis platelet supply in Germany), and the estimate of the number of transmissions of the next major “HIV-like” pathogen to emerge in the future (88 transmissions in the year preceding the implementation of blood-donor screening owing to the continued use of platelet pools in Germany), should be adjusted upward to reflect the 7.2 (rather than 4 or 5) ratio of donor exposures in recipients of pooled versus single-donor platelets (i.e., the ratio that will pertain following the implementation of the patient-centric paradigm [3,87,90]).

In fact, the patient-centric paradigm for clinical transfusion practice [3,87,90] is especially relevant to Germany today, as Germany considers possible alternatives to implementing screening of all donations for WNV RNA in minipools [93]. No cases of WNV infection have been recorded in Germany and donor screening for WNV RNA is not currently performed. However, over the last 3 years (2010-2012), there have been hundreds of cases of WNV infection in European-Union countries (primarily Greece and Italy, but also as close to Germany as Hungary [94]). It is possible that the equivalent of the 1999 (minor) WNV epidemic in New York State has already occurred in Europe (east and south of Germany in 2010-2012 [94]), and that the equivalent of the 2002 (major) WNV epidemic in the US and Canadian plains remains to occur in Europe (including in Germany if the epidemic moves westward, as it did in 2002 in North America [95]). As experience from North America has demonstrated [96], owing to a low viral load dur-
ing the infectious period of acute viremia, only individual or minipool testing of all donations for WNV RNA can protect recipients. Although PR of platelets and plasma would inactivate WNV (should such systems become appropriate for use outside the experimental setting), most recipients of platelets and plasma also receive red blood cells and there is currently no PR system for red blood cells.

Furthermore, until there is a comprehensive system of PR that also encompasses red blood cells, which would permit various cost-savings from the possible discontinuation of other blood safety measures, the cost of PR is bound to be incremental and substantial [97]. The patient-centric paradigm for clinical transfusion practice can meet all of a patient’s transfusion needs while reducing her/his donor exposures at least 2-fold through a combination of patient blood management and multicomponent apheresis [3,90]. After patient blood management and multicomponent apheresis replace the current blood procurement system of whole-blood collections, the reduced number of components collected from allogeneic donors can be tested in minipools for WNV RNA in a precautionary manner (i.e., in anticipation of a future epidemic of WNV in Germany). A 4-week deferral of donors traveling to areas reporting WNV cases may serve as a stop-gap measure until minipool testing of all allogeneic donations is introduced. The risk of WNV transmission by transfusion in Western Europe can thus serve in a constructive manner by helping to reduce overall transmission risk, rather than just the narrow risk of WNV transmission per se. Such will be the case if the effort to confront the risk of WNV transmission brings about changes facilitating the progression of clinical transfusion practice in Western Europe from the component-centric paradigm of the 20th century to the patient-centric paradigm of the 21st century. Several WNV cases of human disease had been reported already in 2008-2009 in northeastern Italy [98,99]. In 2011, 2 new WNV genome sequences from human cases of WNV infection were described. The novel WNV genomes had high nucleotide and amino acid sequence divergence from each other and from the WNV strain circulating in Italy in 2008-2009 [100]. The presence of different WNV strains in a relatively small geographic area is a novel finding that raises the possibility of new WNV strains emerging in the future, which—if mutations occur in the RNA regions targeted by the screening assays—may be undetectable by standard blood-donor screening assays [52]. Although others may advance this possibility as an argument for the introduction of PR of platelets, we sound (once more) the cautionary note that PR for platelets is not ready for implementation [82], while there currently exists no PR system for red blood cells. Therefore, just like the possibility of novel pathogens, the possibility of new viral strains of HIV [52] and WNV [100] is, first and foremost, a reason for reducing donor exposures through widespread implementation of patient blood management and multicomponent apheresis.

In conclusion, since PR is not ready for implementation, the risk of platelet pools differs (and will differ) sufficiently from the risk of single-donor (apheresis) platelets to mandate a revision of the German guidelines for hemotherapy [13] to indicate the difference in risk between these two components; as well as necessitate a planned transition toward an all-apheresis platelet supply. If the safety of PR is established in the future, PR should be performed on an all-apheresis platelet supply to maximize safety; not on the current platelet pools.

Declaration of Interest:
The authors have no conflict of interest to declare.

References:


20. Schriftliche und mündliche Stellungnahme des Paul-Ehrlich-Institut (PEI) zur Anhörung im Sozialpolitischen Ausschuss des Landtag Rheinland-Pfalz, 9. Sitzung am 08.03.2012 (German).
30. Serious hazards of transfusion (SHOT) Annual Reports. Available at: http://www.shot-uk.org
THE CASE FOR AN ALL-APERHESIS PLATELET SUPPLY


82. FDA/CBER. Study Designs (Phases 3 and 4) for Product Development of Human Platelets Using the Cerus INTERCEPT Blood System for Pathogen Inactivation. Meeting of the Blood Products Advisory Committee: November 16, 2009.


84. Jakobs MR, Smith D, Heaton WA, Good CE. Detection of bacterial contamination in prestorage culture-negative apheresis platelets on day of issue with the Pan Genera Detection Test. Transfusion 2011;51:2573-82.


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